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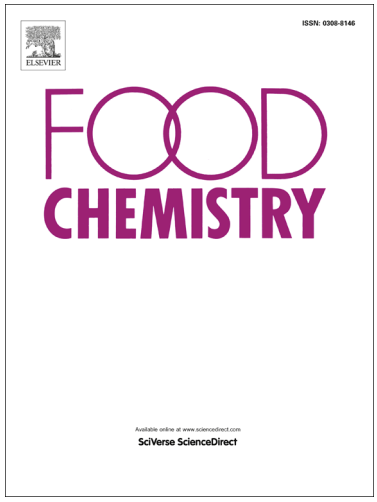

Comparison of the nutritional composition of experimental fermented milk:wheat bulgur blends and commercially available kishk and tarhana products

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Comparison of the nutritional composition of experimental fermented milk:wheat bulgur blends and commercially available kishk and tarhana products.

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Highlights

- Experimental blends of fermented milk and wheat bulgur were prepared.
- Blends with higher fermented milk contents had higher protein, fat and calcium contents.
- Experimental blends compared favourably with similar commercially available products.

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ABSTRACT

Dried, fermented blends of dairy products and cereals, such as kishk and tarhana, are foodstuffs traditionally consumed in many regions as they possess good nutritional qualities and extended storage stability. This study examined the nutritional composition of kishk or tarhana type products and compared with experimental blends of fermented milk and wheat bulgur containing 60% to 80% milk. The blends with higher milk contents had levels of protein (18.9%) and fat (5.8%) at the concentrations specified in fortified blended foods as outlined by the World Food Program. Higher milk contents were also associated with higher contents of calcium (323.2mg/100g), phosphorus (335.3mg/100g), vitamin A (486.7µg/100g) and α-tocopherol (174.5 µg/100g). The nutritional content of the experimental fermented milk:wheat bulgur blends compared favourably with that of the commercial samples. These blends may be suitable as base products, to be fortified with micronutrients, for the development of fortified blended foods (FBFs) for humanitarian distribution.

Keywords: Kishk; Tarhana; Fermented milk; Wheat bulgur; Mineral; Nutritional value.

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1. Introduction

Kishk is a fermented, dried blend of milk (whole, skim milk or buttermilk) and cereal (usually bulgur) which is consumed in many areas of the Middle East, Africa and Asia (Tamime & O'Connor, 1995). The combination of milk or yoghurt and cereal enhances the essential amino acid profile of the end product (Sarwar, 1997). Additional health promoting benefits may also be conferred through combinations of dairy and cereal (Mahmoudi, 2014). Fermentation may improve the nutritional value of dairy-cereal blends by reducing the content of anti-nutrients, such as phytic acid, thereby increasing mineral bioavailability (Poutanen, Flander & Katina, 2009); and also by increasing the content of bioactive compounds (Rahmawati & Suntornsuk, 2016). Kishk and similar products, such as tarhana, are often prepared in a domestic setting; hence, there is large variation in the composition of these products (Maskan & İbanoglu, 2002; Sadrizadeh et al., 2018). In general, kishk and tarhana have a dairy content ranging from 60-80% w/w (Robinson & Cadena 1978; Tamime & O'Connor, 1995) and tarhana is often flavoured by the incorporation of vegetables or spices (Georgala, 2013). The composition, nutritional properties and sensory quality of kishk and tarhana can be modified by changing the type of cereal, starter culture and the proportion of dairy ingredients used in the formulation (Gadallah & Hassan, 2017; Demirci, Ibrahim, Ozalp & Tirpanci Sivri, 2018).

Fermented blends of dairy and cereal have the potential to be fortified with selected nutrients and hence developed as fortified blended foods (FBFs) to meet the nutritional needs of young children and malnourished individuals in low income countries. Existing FBFs, supplied by the World Food Program, are composed of cereal (maize, wheat or rice), soy or other legume and a micronutrient premix to meet the requirements for essential vitamins and minerals. Corn-soy blend (CSB) has been shown to have limitations as evidenced by a retrospective review of emergency supplementary feeding programmes which found sub-optimal recovery

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rates in children (Navarro-Colarado, 2007). In comparison to CSB, lipid nutrient supplements were more effective at increasing the fat free mass index of children with moderate acute malnutrition (MAM) following a 12 week nutritional intervention (Fabiansen et al., 2017). The intake of animal-source foods, including milk, has been positively associated with weight and height gain in children with poor diets (Marquis, Habicht, Lanata, Black & Ramussen, 1997; Smith, Earland, Bhatia, Heywood & Singleton, 1993). Improved MAM recovery rates and increased growth were observed in children that received a ready to use therapeutic food (RUSF) with whey protein concentrate compared with those that received a RUSF with soy protein (Stobaugh et al., 2016). A FBF formulation (SUPER CEREAL *plus*-Corn soya blend) containing added skim milk powder, which results in a higher energy density and improved micronutrient content, has been developed in recent years.

The objective of this research was to compare the nutritional content of five commercial tarhana (T1-T5) and three kishk (K1-K3) products with that of an experimental fermented milk:bulgur wheat blend (FMB) produced as described in Shevade et al. (2018a). The formulation of the FMB was modelled on kishk and they contained 60, 65, 70, 75 or 80% fermented milk derived from Irish skim milk powder (SMP) and buttermilk powder (BMP). The fact that dried fermented cereal/dairy products are traditionally consumed in areas of the world that are susceptible to humanitarian emergencies make our FMB particularly suited for fortification to produce novel FBFs.

2. Materials and methods

2.1 Materials

All chemicals were purchased from Sigma-Aldrich Ireland Limited (Wicklow, Ireland) unless otherwise stated. All solvents were of certified HPLC grade. Commercial brands of Tarhana

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(T1-T5) and Kishk (K1-K3) powders were procured from Syria, Greece, Turkey and Lebanon (for ingredients of each product see appendix 1). Samples were stored in airtight containers in the dark. BMP (protein 33%, fat 7%, lactose 46%, lactic acid 0.23%) was kindly supplied by Glanbia Ingredients plc. (Ballyragget, Co. Kilkenny, Ireland). Extra low-heat skim milk powder (protein 38.43%, fat 0.89%, lactose 46.2%, lactic acid 0.04%) was manufactured using a pilot-scale NIRO Tall-Form Dryer in Moorepark Technology Limited (Cork, Ireland), as described by Lin et al. (2016). Cream (fat 37%) and wheat bulgur were purchased from a local retail store.

2.2 Methods

2.2.1 Production of experimental fermented milk:bulgur blends (FMB)

Three separate batches of the FMB were produced as detailed in Shevade et al., (2018a). Reconstituted milk was prepared by dispersing BMP and low-heat skim milk powder in distilled water to 7.0 and 8.9% total solids, respectively; cream was then added to standardise the fat content to 2%. The reconstituted milk was heat treated at 95°C for 2.5 minutes, homogenised, cooled to 42°C, inoculated with *Streptococcus thermophilus* and *Lactobacillus delbrueckii subsp bulgaricus*, at 42°C until the pH dropped to 4.6, cooled to 15°C while stirring, and stored overnight at 4°C. The fermented milk and bulgur were blended at weight ratios of 80:20, 75:25, 70:30, 65:35 and 60:40, and were incubated for 24 hours at 35°C (Heratherm Incubator, ThermoFisher Scientific, Waltham, MA, USA) to allow hydration of the parboiled bulgur. Following fermentation, the products were spread over a drying tray and dried to 4%-7% moisture content (Excalibur® dehydrator, Sacromento, CA, USA). The dried products were milled to 1mm (Ultracentrifugal Mill ZM 200 fitted with a trapezoidal 1mm ring sieve; Retsch Technology GmbH, Haan, Germany) and stored in sealed plastic bags at room temperature, in the dark. The FMB powders formulated with 80, 75, 70, 65 and

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60 % (w/w) fermented milk were denoted as FMB80, FMB75, FMB70, FMB65 and FMB60, respectively.

2.2.2 Compositional analysis of Tarhana and Kishk powders

Tarhana and kishk powders were milled to a mesh size of ~1mm prior to analysis. All powders were analysed in triplicate for protein by Kjeldahl (AOAC, 1995), fat by solvent extraction and distillation using Rose-Gottlieb (IDF, 2010). Starch, total dietary fibre, lactose, and lactic acid were determined using the Megazyme K-TSHK 09/15, KTDFR 12/15, K-LACGAR, and K-DLATE kits, respectively (Megazyme International Ireland, Bray Business Park, Bray, Co. Wicklow, Ireland). NaCl was measured by potentiometric determination of chloride (Conroy, 2018).

2.2.3 Nutritional Analysis

Quantification of calcium, iron, zinc and magnesium was conducted based on AOAC method 999.11 (Jorhem & Engman, 2000). Briefly, each sample (1g) was dry-ashed by placing at 500°C in a muffle furnace for 4 hrs. The ashed samples were dissolved in 3M HCl (40mL), a few drops of HNO_3 were added and the samples were boiled. Each sample was then adjusted to a final volume of 50mL and concentrations of calcium, magnesium, iron and zinc were analysed using a flame atomic absorption spectrophotometer (Varian, SpectrAA-600) at wavelengths of 422.7nm, 285.2nm, 248.8nm and 213.9nm, respectively.

Fatty acid analysis was carried out using a direct derivatisation method as described in Castro-Gómez, Fontecha & Rodríguez-Alcalá (2014). Briefly, the sample (0.1g) was placed in a Pyrex tube and 1M methanolic H_2SO_4 (3mL) was added. Samples were heated at 80ºC for 1.5 hours and then cooled on ice. Hexane (1mL) was added, samples were vortexed and 6% (w/v) Na_2CO_3 (7.5mL) was added to each tube. The samples were then centrifuged

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(3,500rpm for 10mins) and the upper layer was transferred to an eppendorf and stored at -20ºC prior to analysis. The fatty acid methyl esters (FAME) analysis was conducted by capillary-column gas liquid chromatography (GC) as previously described by Ryan et al. (2007). Data were presented as the percentage of each fatty acid relative to the total fatty acid content of the sample.

Phytic acid was determined using a Megazyme kit (K-PHYT; Co. Wicklow, Ireland). Mixed linkage β-glucan was measured in each of the samples using Megazyme kit (K-BGLU; Co.Wicklow, Ireland).

For the determination of α-tocopherol, vitamin A and carotenoids, extracts of each sample were prepared according to the method described in Menkir, Gedil, Tanumihardjo, Adepoju & Bossey (2014). The samples (0.5 g) were mixed with ethanol (6mL) and heated to 85°C for 10 min, 80% KOH (0.5 mL) was added and samples were held at 85°C for a further 10 min. The extraction was carried out using hexane which was subsequently removed by solvent evaporation (miVac solvent evaporator, Genevac Ltd, Suffolk, UK). α-Tocopherol, vitamin A and carotenoid concentrations were quantified by reverse-phase HPLC. For α-tocopherol and vitamin A analysis, the HPLC system consisted of two LC-20ADXR pumps, an SIL-30AC autosampler, a CTO-20AC column oven, an SPD-M30A photodiode array (PDA) detector and an RF-20AXS fluorescence detector (Shimadzu Corp., Kyoto, Japan). The column was an Ascentis Express C18 (100 x 4.6mm i.d., 2.7µm; Sigma-Aldrich Ireland Limited, Wicklow, Ireland). The extracts were dissolved in 200µL methanol and the injection volume was 20µL. The mobile phase was methanol:water (97:3) and elution was isocratic at a flow rate of 1mL/min. Column temperature was maintained at 30°C. The PDA detector was set to scan from 200-500nm and spectra were extracted at 325nm for vitamin A analysis. The fluorescence detector excitation and emission wavelengths were set at 293 and

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326nm, respectively, for α-tocopherol analysis. Data were recorded and processed using LabSolutions Lite software (Shimadzu Corp.). For carotenoid analysis, samples were dissolved in 200µL methanol:ethyl acetate (4:1, v:v) and separated at 33°C by gradient elution on a YMC C30 reversed phase column, as detailed in Pugliese et al. (2013).

2.2.4 Antioxidant Activity and total phenol content

An extract of each sample was prepared by placing sample (0.3g) in a glass tube and adding 70% aqueous acetone (10mL). The samples were vortexed well and incubated at room temperature for 30 minutes with intermittent vortexing. Following incubation, the samples were centrifuged at 15,000 x g for twenty minutes and the supernatant was isolated. Extracts were analysed for their total phenol content (TPC) using the method outlined in Park et al. (2018). The ferric reducing antioxidant power (FRAP) assay and the oxygen radical absorbance capacity (ORAC) assay were carried out as previously detailed in Sae-leaw, O'Callaghan, Benjakul & O'Brien (2016).

2.3 Statistical Analysis

Data are expressed as the mean ± SD. Each brand of kishk and tarhana was analysed in triplicate. Statistical differences between kishk and tarhana commercial products were determined by ANOVA followed by Tukey's multiple comparison test. Statistical differences between the different formulations of experimental FMBs were also determined by ANOVA followed by Tukey's multiple comparison test. One-way ANOVA followed by Tukey's multiple comparisons test was performed using GraphPad Prism version 7.00 for Windows, GraphPad Software, La Jolla California USA, www.graphpad.com. A value of $P<0.05$ was deemed to be significant.

3. Results and discussion

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3.1 Gross nutritional composition

The kishk samples had significantly (P<0.05) higher protein contents than the tarhana samples; the highest protein content was in the experimental blend, FMB80, which contained 80% fermented milk (Table 1). An increase in the fermented milk content (60-80%) correlated with an increase in the protein content (15.2-18.9 %) of the FMBs. The protein content of the dried, fermented milk (FM) and wheat (dry matter) were 33.4% (w/w) and 12.0% (w/w), respectively (Shevade et al., 2018b). The blends with the highest fermented milk contents, FMB 80, FMB 75 and FMB 70, had protein levels sufficient to meet requirements outlined by the WFP for wheat soya blend (WSB) which should contain a minimum of 16% protein (WFP 2015). In general, the fat content of the commercial samples and the experimental blends was below 6% with the exception of two of the kishk products (K1 and K2) which contained 15.1% and 9.6% fat, respectively (Table 1). The fat content of the FM (dry matter) and wheat (dry matter), used in the preparation of FMB, were 12.5% (w/w) and 2.4% (w/w), respectively (Shevade et al., 2018b). In existing FBF products, the fat content of CSB and WSB is increased by the addition of soya oil (WFP, 2014), at a concentration of 4% (w/w). Salt was higher (0.4- 5.4%) in the commercial tarhana and kishk products (Table 1) but did not exceed 0.7% in the experimental FMBs. Salt is frequently added to limit microbial growth and add flavour however, sodium should be minimised in products designed for malnourished children (Golden, 2009). The experimental FMBs were found to be microbiologically safe in the absence of any added salt (data not shown). The content of lactose in the experimental blends increased as the fermented milk content was increased (Table 1) and was highest in the blend with the highest fermented milk content (14.9%). The lactose content of the FM was 7.43% (w/w). Lactose is the primary energy source in human breast milk and may have a number of benefits for malnourished children (Grenov et al., 2016). It has been recommended that lactose should be reduced in the diets of

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malnourished children with diarrhoea but recent evidence suggests that the benefits of lactose should be further investigated (Grenov et al., 2016).

3.2 Mineral content of commercial and laboratory scale products

The calcium content of the kishk samples, K1-K3 (109.3-230.9mg/100mL) differed significantly (P<0.05) but all kishk samples contained significantly higher (P<0.05) calcium contents than the tarhana products which had calcium contents ranging from 14.7 to 52.6mg/100g (Table 2). The highest calcium content (323.3mg/100g) was in the experimental blend containing 80% fermented milk (Table 2) and the content of calcium decreased significantly (P<0.05), in correlation with the decreasing fermented milk content of the samples, to 145.2mg/100g in the product containing 60% fermented milk (Table 2).

The mean contents of magnesium, iron, zinc and phosphorus of the Kishk powders were approximately 2-fold higher than that of the tarhana (Table 2). The levels of magnesium were similar in each of the experimental blends and were higher than the levels in tarhana but lower than the levels in kishk (Table 2). The experimental FMBs had a lower content of iron than either the kishk or the tarhana samples and the iron content did not vary significantly with the changing weight ratio of fermented milk to bulgur (Table 2). Similarly, the zinc content did not significantly differ in the various experimental samples. The content of phosphorus (271.7-335.3mg/100g) in the experimental blends increased significantly (P<0.05) with the increasing fermented milk content (Table 2).

An investigation of 25 varieties of Lebanese kishk, conducted by Tamime, Barclay, McNulty & O'Connor (1999), found calcium, magnesium, zinc and iron contents ranging from 138.6 to 340.0 mg/ 100g; 94.9 to 167.8 mg/100g; 2.5 to 4.0mg/100g and 3.7 to 9.5mg/100g, respectively; therefore, the kishk samples analysed in the present study (Table 2) were within the range previously reported for similar type products. Iron, zinc and calcium are regarded

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as problem essential nutrients by the WHO and an analysis of 57 complementary food products, currently on the market, revealed that only 4% contained sufficient iron, zinc and calcium to meet the dietary requirements of breast-fed infants aged 9-11 months (Gibbs et al., 2011). The experimental FMB would require fortification with each of these minerals in order to comply with the specifications of the WFP which recommends that fortified blended foods (Supercereal plus) should contain calcium, iron and zinc at concentrations of 362mg/100g, 6.5mg/100g and 5mg/100g, respectively (WFP, 2015).

3.3 Fat-soluble vitamins and carotenoids

The content of α-tocopherol was significantly (P<0.05) higher in the tarhana products which contained red pepper (686.7µg/100g- 1479.9µg/100g) in comparison with the samples which did not contain red pepper (Table 2). The FMB contained 105.8 to 174.5µg/100g α-tocopherol. Vitamin A was below the level of detection in the majority of the tarhana and kishk samples with the exception of samples T1 and K2 (Table 2); it is possible that vitamin A may have deteriorated during storage of these products. Vitamin A is susceptible to oxidation and has been shown to decrease by up to 85% following 3 months storage in fortified wheat flour (Hemery et al., 2018). α-Tocopherol and vitamin A contents of the FMB increased significantly (P<0.05) as the weight proportion of fermented milk was increased from 60 to 80% (Table 2). FMB80 had a vitamin A content (Table 2) of just below 50% of that recommended by the WFP (1,038µg/100g). In a previous analysis of 25 varieties of kishk, the maximum vitamin A content was 102.7µg/g and the maximum α-tocopherol content was 332.0µg/100g (Tamime et al., 1999).

Neither the commercial samples nor the experimental FMB were a substantial source of carotenoids (Table 1). The highest carotenoid contents were in the tarhana samples which had red pepper listed as an added ingredient, and these samples had significantly (P<0.05)

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higher contents of lutein and β-carotene and were also the only samples with detectable levels of zeaxanthin and capsanthin, a carotenoid which is unique to the capsium family (Jung et al., 2015). In the experimental FMB bulgur products, the lutein content increased significantly (P<0.05) as the bulgur content increased and β-carotene increased significantly (P<0.05) with increasing the content of fermented milk. There are currently no recommendations for the inclusion of carotenoids in FBFs. However, carotenoids can act as antioxidants within the food system to prolong shelf-life (Siwach, Tokas & Seth, 2016) and *in-vivo* to provide potential health benefits (Jomova & Valko, 2013). In addition, certain carotenoids, particularly β-carotene, possess pro-vitamin A activity and are the primary source of vitamin A amongst populations with limited availability of animal foods (Weber, & Grune, 2012).

3.4 Fatty acid profile

The fatty acid data were expressed as a percentage of the total fatty acid content (Table 3). The predominant fatty acid in each of the kishk samples was C16:0, palmitic acid (30.9-40.5%), followed by C18:1, oleic acid (10.6-18.8%). There was large variation in the proportion of the essential fatty acid (EFA), linoleic acid (C18:2), between the kishk samples (3.8% to 24.3%); a similar range (2.18% to 20.89%) was previously found in a survey of 25 Lebanese commercial kishk samples (Tamime et al., 1999). There was no α-linolenic acid (C18:3) detected in the tarhana samples but it is possible it may have degraded during storage. In general, tarhana samples had higher proportions of linoleic acid (22.5-49.6%) than kishk samples (3.8-24.3%), with higher proportions of linoleic acid particularly evident in the tarhana samples containing red pepper (T3, T4 and T5). The predominant fatty acid in wheat is linoleic acid, at approximately 50% of the total fatty acid content (Hidalgo, Brandolini & Ratti, 2009). The higher content of linoleic acid in tarhana is indicative of a higher wheat content in comparison with the kishk samples. Paprika, the ground, dried form

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of red peppers has a linoleic content of approximately 69% of the total fatty acid content (Pérez-Gálvez, et al., 1999) and this could result in the higher percentage of linoleic acid observed in the tarhana samples containing red pepper. The samples containing red pepper also had a lower percentage of short and medium chain fatty acids (C4:0- C14:0) and improved ratios of unsaturated fatty acids to saturated fatty acids (1.59-2.25 %). The proportion of saturated fatty acids, and oleic acid, in the experimental fermented milk bulgur products, increased on increasing the fermented milk content (Table 3). The FMB products had higher percentages of linolenic acid than kishk or tarhana (Table 3).

An adequate dietary intake of EFA (linoleic and α-linolenic acid) is required for optimal functioning of the immune system and the brain (Chang, Ke & Chen, 2009). A recent study demonstrated that the proportion of polyunsaturated fatty acids (PUFA), including EFA, was lower in the blood of children with severe acute malnutrition in comparison to healthy children and suggested that dietary interventions should place increased emphasis on the intake of PUFA (Babirekere-Iriso et al., 2016). Currently, there are no specifications for the concentration of essential fatty acids required in FBFs but a recommendation of 4.5% energy in the form of n-6 PUFA and 0.5% energy in the form of n-3 PUFA for undernourished children has been proposed (Golden, 2009). As observed for the tarhana samples (Table 3), the inclusion of fruit/vegetable may improve the PUFA content. A high content of unsaturated fatty acids may increase oxidation in the end-product. Therefore, studies on the optimal levels of fatty acids to be incorporated into FBFs and their interaction with other ingredients in the formulation are required.

3.5 β-D-(1-3, (1-4)-glucan

The mean β-glucan content of kishk samples (113.3-344.8mg/100g) was significantly higher than that of the tarhana samples (38.2-127.2mg/100g). β-glucan is a cell wall carbohydrate

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and the lower content of tarhana samples (T2 –T5) could therefore result from the use of a refined wheat flour, rather than whole grain, during formulation. In a previous study, the β-glucan content of kishk type products ranged from 140 to 610mg/100g (Tamime et al., 1999). The experimental FMB, which were prepared using whole grain bulgur, had β-glucan levels which ranged from 169.2 to 238.7mg/100g (Table 4). The β-glucan content of the experimental blends increased in association with an increasing bulgur content, up to a bulgur content of 35%. Potential health benefits which have been linked to the consumption of β-glucans include the lowering of serum cholesterol levels (Zhu et al., 2015) and an enhanced regulation of the response to glucose and insulin in diabetics (Cavallero, Empilli, Brighenti & Stanca, 2002). The content of β-glucan in wheat, barley and oats were reported to be in the ranges of 1%, 3–11% and 3–7%, respectively (Skendi, Biliaderis, Lazaridou & Izydorczyk, 2003), hence there is the potential to increase the β-glucan content of products modelled on kishk by substituting oat or barley for bulgur. Barley flours have previously been used to produce tarhana with a β-glucan content approximately 10-fold higher than that of a wheat tarhana (Erkan et al., 2006). The authors noted that the fermentation process resulted in some loss of β-glucan (Erkan et al. 2006).

3.6 Phytic acid

The quantity of phytic acid in the tarhana products (55.8-132.5mg/100g) was approximately 5-fold lower than in either the kishk (426.7-680.6mg/100g) or the experimental FMB (369.9-465.4mg/100g). The tarhana may have been produced using refined flour which has a lower phytic acid content than wholemeal flour (Febles, Arias, Hardisson, Rodriguez-Alvarez & Sierra, 2002). There was no significant difference in the phytic acid content of the experimental samples with various bulgur content (Table 4). Phytic acid is an anti-nutrient which chelates calcium, iron and zinc, and thereby reduces their absorption *in-vivo* (Raboy,

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2003). Magala, Kohajdová & Karovičová (2015) showed that the phytic acid content of a cereal beverage was reduced to 80% of the original phytic acid content following a 24 hour fermentation, using lactic acid bacteria. Roos et al. (2013) analysed the content of phytic acid in food aid products and found that fortified blended foods contained phytic acid at concentrations ranging from 142 to 1,000 mg/100g and the majority of the products analysed did not meet the recommended phytate:mineral molar ratios. The authors recommended that upper limits for the phytic acid content of processed foods should be established (Roos et al., 2013). The phytic acid content of future FBF products could be reduced by dehulling the bulgur or other cereal, by the addition of phytase or by incorporating a fermentation step during the production of the FBF.

3.7 Total Phenolic content (TPC), ferric reducing antioxidant power (FRAP) and oxygen radical absorbance capacity (ORAC) assays

The TPC in the kishk samples ranged from 47.3 to 62.1 mg gallic acid equivalents (GAE)/100g (Table 5). The tarhana products which contained red pepper as an added ingredient (T3, T4 and T5) had significantly ($P<0.05$) higher TPC than either the kishk samples or the tarhana products which did not contain red pepper. The highest FRAP and ORAC activities were also found in the tarhana samples which contained red pepper (Table 5). The highest ORAC activity was in sample T4 which had the highest total phenol content and the highest FRAP value was in sample T5 (Table 5). The total phenol content of the experimental FMB samples ranged from 41.2 to 52.5mg GAE /100g (Table 5) and there was no evident association between the bulgur content of the samples and their TPC. Phenolic acids are primarily found in plant foods but have also been detected in milk (Vázquez et al., 2015). There was no significant difference between the antioxidant activity of the different laboratory-made samples as assessed by the FRAP and the ORAC assays (Table 5).

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A wheat tarhana was previously found to have a free phenolic content of 122.5mg GAE/100g and the phenolic content and antioxidant activity were significantly increased by partial substitution of wheat with oats (Kilci & Gocmen 2014). Polyphenols may form a complex with proteins and minerals which can affect their bioaccessibility (Jakobek, 2015; Ozda, Capanoglu & Altay, 2013) and it has been recommended that phenolics and other anti-nutrients should be reduced or eliminated in complementary foods (FAO, 2013). However, phenolic compounds limit lipid oxidation (Siriamornpun, Tang, Khawanit & Kaewseejan, 2016) and thereby, reduce rancidity and enhance storage stability. Malnutrition can cause an increase in oxidative stress which could be alleviated by the consumption of a diet rich in antioxidants (Ghone et al., 2013).

4. Conclusion

Overall, the kishk products had a higher mineral and β-glucan content than the tarhana products but they also had a higher phytic acid content. The inclusion of red pepper as an ingredient in tarhana increased the α-tocopherol and carotenoid contents, antioxidant activity and improved the fatty acid profile. The nutrient content of the experimental blends was more similar to the kishk products than the tarhana products. As the fermented milk content of the experimental blends was increased there was a corresponding increase in protein, fat, calcium, phosphorus, vitamin A, α-tocopherol and essential fatty acids and a decrease in β-glucan. There was no significant difference in the phytic acid content or the antioxidant activity of the various experimental blends. This study demonstrated the nutritional benefit of increasing the content of dairy in FBF formulations however, further study is necessary to determine if the potential benefits to human health outweigh the costs involved in increasing the quantity of dairy used in the preparation of FBFs. Data obtained for the tarhana products

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containing red pepper illustrate the possibility of developing products with enhanced storage stability and nutrient content by the incorporation of fruit or vegetable derived ingredients.

Conflict of interest statement

The authors declare that there was no conflict of interest.

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Table 1

Compositional parameters of tarhana and kishk powders and experimental fermented milk:bulgur blends (FMB)

	Commercial Tarhana					Commercial Kishk			Experimental FMB				
	T1	T2	T3	T4	T5	K1	K2	K3	FMB80	FMB75	FBM70	FMB65	FMB60
Protein (%)	$11.7\pm0.1^{b\text{-}h}$	$11.0\pm0.0^{a,c\text{-}g}$	$8.7\pm0.1^{a,b,d\text{-}h}$	$9.1\pm0.1^{a\text{-}c,f\text{-}h}$	$9.4\pm0.2^{a\text{-}c,f\text{-}h}$	$8.2\pm0.1^{a\text{-}e,g,h}$	$17.5\pm0.1^{a\text{-}f,h}$	$11.1\pm0.1^{a,c\text{-}g}$	$18.9\pm0.3^{B\text{-}E}$	$17.9\pm0.4^{A,C\text{-}E}$	$16.7\pm0.1^{A,B,D,E}$	$15.9\pm0.4^{A\text{-}C,E}$	$15.2\pm0.4^{A\text{-}D}$
Fat (%)	$5.4\pm0.2^{b,e\text{-}h}$	$2.7\pm0.1^{a,f,g}$	$4.0\pm0.6^{f,g}$	$4.2\pm1.5^{f,g,h}$	$3.4\pm1.0^{a,f,g}$	$15.1\pm0.8^{a\text{-}e,g,h}$	$9.6\pm0.2^{a\text{-}f,h}$	$2.7\pm0.6^{a,d,f,g}$	$5.8\pm0.4^{B\text{-}E}$	$5.1\pm0.3^{A,D,E}$	$4.6\pm0.3^{A,E}$	$4.1\pm0.2^{A,B}$	$3.7\pm0.2^{A\text{-}C}$
Starch (%)	$66.8\pm4.8^{b,f,g}$	$57.0\pm2.4^{a,f,g}$	$62.0\pm2.4^{f,g}$	$63.5\pm5.0^{f,g}$	$62.8\pm0.2^{f,g}$	$40.0\pm3.2^{a\text{-}e,h}$	$42.0\pm2.3^{a\text{-}e,h}$	$58.3\pm1.1^{f,g}$	$38.9\pm1.2^{C\text{-}E}$	$42.3\pm1.7^{C\text{-}E}$	$47.1\pm2.4^{A,B,E}$	$48.8\pm1.1^{A,B}$	$52.0\pm2.5^{A\text{-}C}$
Dietary fibre (%)	$3.3\pm0.5^{f\text{-}h}$	$3.0\pm0.6^{f\text{-}h}$	$3.0\pm0.1^{e\text{-}h}$	$3.5\pm0.4^{f\text{-}h}$	$4.2\pm0.2^{c,f\text{-}h}$	$6.1\pm0.7^{a\text{-}e,g,h}$	$7.3\pm0.2^{a\text{-}f,h}$	$8.5\pm0.2^{a\text{-}g}$	ND	ND	ND	ND	ND
Lactose (%)	$1.1\pm0.1^{c\text{-}h}$	$1.0\pm0.1^{c\text{-}h}$	$1.5\pm0.1^{a,b,d\text{-}h}$	$0.2\pm0.0^{a\text{-}c,g}$	$0.5\pm0.0^{a\text{-}c,g}$	$0.1\pm0.0^{a\text{-}c,g}$	$3.4\pm0.2^{a\text{-}f,h}$	$0.5\pm0.0^{a\text{-}c,g}$	$14.9\pm0.9^{C\text{-}E}$	$13.7\pm0.9^{C\text{-}E}$	$10.6\pm1.1^{A,B}$	$10.2\pm0.7^{A,B}$	$8.6\pm0.7^{A,B}$
Salt (%)	$2.3\pm0.2^{b\text{-}h}$	$4.8\pm0.1^{a,c\text{-}h}$	$1.9\pm0.0^{a,b,d\text{-}h}$	$0.4\pm0.0^{a\text{-}c,e\text{-}h}$	$1.5\pm0.0^{a\text{-}d,f\text{-}h}$	$2.7\pm0.1^{a\text{-}e,g,h}$	$5.4\pm0.1^{a\text{-}f,h}$	$4.5\pm0.0^{a\text{-}g}$	$0.7\pm0.0^{B\text{-}E}$	$0.6\pm0.0^{A,C\text{-}E}$	$0.5\pm0.0^{A,B,D,E}$	$0.5\pm0.0^{A\text{-}C,E}$	$0.4\pm0.0^{A\text{-}D}$
Lactic Acid (%)	$0.6\pm0.1^{c\text{-}h}$	$0.6\pm0.0^{c\text{-}h}$	$0.9\pm0.1^{a,b,d\text{-}h}$	$1.8\pm0.1^{a\text{-}c,e\text{-}h}$	$0.1\pm0.0^{a\text{-}d,f\text{-}h}$	$2.7\pm0.1^{a\text{-}e,g,h}$	$1.8\pm0.0^{a\text{-}f,h}$	$2.4\pm0.2^{a\text{-}g}$	$4.2\pm0.2^{D,E}$	$3.8\pm0.2^{D,E}$	$3.6\pm0.4^{D,E}$	$3.0\pm0.3^{A\text{-}C}$	$2.7\pm0.3^{A\text{-}C}$

ND: Not determined

The presented data are the means of three replicate samples for different retail brands of tarhana (T1-T5) or kishk (K1-K3) powders and different experimental FMB consisting of 20-40% bulgur and 80-60% fermented milk (FMB80-FMB60)

$^{a\text{-}h}$ indicates significant difference ($P<0.05$, ANOVA followed by Tukey's test) between kishk or tarhana samples (^{a} is T1, ^{b} is T2 etc.)

$^{A\text{-}E}$ indicates significant difference ($P<0.05$, ANOVA followed by Tukey's test) between experimental FMB samples (^{A} is T1, ^{B} is T2 etc.)

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Table 2
Mineral, α-tocopherol, vitamin A and carotenoid content of retail brands of tarhana and kishk powders and experimental fermented milk:bulgur blends (FMB)

	Commercial Tarhana					Commercial Kishk			Experimental FMB				
	T1	T2	T3	T4	T5	K1	K2	K3	FMB80	FMB75	FMB70	FMB65	FMB60
Calcium (mg/100g)	32.1±3.9^{f-h}	52.6±7.5^{f-h}	27.2±2.4^{f-h}	14.7±3.4^{f-h}	37.2±1.9^{f-h}	230.9±29.5$^{a-e,g,h}$	173.5±19.2$^{a-f,h}$	109.3±13.6^{a-g}	323.2±3.2^{B-E}	280.6±15.5$^{A,C-E}$	226.5±2.3^{A,B,D,E}	177.7±1.8$^{A-C,E}$	145.2±5.6^{A-D}
Magnesiun (mg/100g)	50.3±0.3$^{b,c,f-h}$	33.8±0.2$^{a,d-h}$	40.8±1.2$^{a,d-h}$	48.5±6.3$^{b,c,f-h}$	52.1±1.1$^{b,c,f-h}$	75.4±1.9$^{a-e,g,h}$	97.7±1.7$^{a-f,h}$	102.5±1.0^{a-g}	76.6±0.6^{E}	82.7±4.2^{E}	77.2±1.3	78.4±1.0	74.6±4.5^{A,B}
Iron (mg/100g)	1.7±0.2^{b-h}	3.3±0.1^{a,g,h}	3.3±0.1^{a,d,g,h}	3.0±0.3^{a,c,g,h}	3.2±0.4^{a,g,h}	2.8±0.3^{a,g,h}	4.8±0.3$^{a-f,h}$	7.3±0.6^{a-g}	1.5±0.1^{B-E}	1.7±0.1^{A}	1.7±0.2^{A}	1.8±0.1^{A}	1.8±0.1^{A}
Zinc (mg/100g)	1.3±0.2^{f-h}	1.1±0.3^{f-h}	1.6±0.1^{f,g}	1.1±0.2^{f-h}	1.1±0.2^{f-h}	3.0±0.3^{a-e}	3.1±0.3^{a-e}	2.3±0.5^{a,b,d,e}	2.2±0.4	2.0±0.1	1.9±0.1	1.9±0.1	2.0±0.2
Phosphorus (mg/100g)	121.4±19.3^{f,g}	87.9±27.6^{f-h}	107.9±21.5^{f-h}	131.7±22.0^{f,g}	117.4±25.7^{f,g}	359.1±29.7$^{a-e,g,h}$	257.2±35.8$^{a-e,g,h}$	181.1±19.3^{b,c,f,g}	335.3±4.0^{D,E}	318.4±16.7	299.2±22.3	289.4±23.6^{A}	271.7±20.6^{A}
Vitamin A (μg/100g)	137.0±9.7	nd	nd	nd	nd	nd	91.4±13.6	nd	486.7±14.5^{C-E}	387.6±65.7	286.5±61.1^{A}	224.1±51.4^{A}	209.5±38.3^{A}
α-Tocopherol (μg/100g)	62.5±3.0^{c-e}	36.4±4.7^{c-e}	1479.9±208.6$^{a,b,d,f-h}$	686.7±40.6$^{a-c,e-h}$	1469.3±41.6$^{a,b,d,f-h}$	12.9±2.2^{c-e}	31.1±0.7^{c-e}	28.0±2.5^{c-e}	174.5±19.0^{D,E}	148.3±16.2^{E}	128.0±26.3^{E}	113.1±17.3^{A}	105.8±4.3^{A-C}
β-Carotene (μg/100g)	0.4±0.0^{c-e}	0.3±0.0^{c-e}	40.5±0.7^{a,b,d,e}	16.1±0.4^{a,b,c,e}	28.2±0.5^{a,b,c,d}	nd	nd	nd	2.2±0.4^{E}	2.0±0.5	1.8±0.3	1.4±0.2	1.4±0.4^{A}
Lutein (μg/100g)	54.3±6.8^{b,d,e,f,h}	10.8±0.3$^{a,c-e}$	60.4±0.3$^{b,d-f,h}$	75.6±0.6$^{a-c,e,f,h}$	142.9±0.4$^{a-d,f,h}$	2.2±0.1$^{a,c-e,h}$	nd	4.5±0.3$^{a,c-e}$	18.1±3.5^{E}	16.4±7.0^{E}	23.0±2.5	23.5±2.1	29.2±6.8^{A,B}
Zeaxanthin (μg/100g)	nd	nd	32.1±0.9^{e}	30.6±1.3^{e}	49.4±0.8^{c,d}	nd	nd	nd	nd	nd	nd	nd	nd
Capsanthin (μg/100g)	nd	nd	70.2±0.6^{d}	45.2±8.5^{c,e}	85.0±9.2^{d}	nd	nd	nd	nd	nd	nd	nd	nd

nd: Not Detected

The presented data are the means of three replicate samples for different retail brands of tarhana (T1-T5) or kishk (K1-K3) powders and experimental FMB consisting of 20-40% bulgur and 80-60% fermented milk (FMB80-FMB60)
$^{a-h}$ indicates significant difference (P<0.05, ANOVA followed by Tukey's test) between kishk or tarhana samples (^{a} is T1, ^{b} is T2 etc.)
$^{A-E}$ indicates significant difference (P<0.05, ANOVA followed by Tukey's test) between experimental FMB samples (^{A} is T1, ^{B} is T2 etc.)

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Table 3
Fatty acid profile (% Total fatty acid content) of retail brands of tarhana and kishk powders and experimental fermented milk:bulgur blends (FMB)

	Tarhana					Kishk			Experimental FMB				
	T1	T2	T3	T4	T5	K1	K2	K3	FMB80	FMB75	FMB70	FMB65	FMB60
C4:0	4.4	2.0	0.2	0.2	0.0	1.5	0.6	0.4	0.9	1.0	1.2	0.5	0.7
C6:0	3.5	1.9	0.0	0.0	0.0	2.2	2.0	0.9	1.2	1.3	1.4	1.0	1.1
C8:0	1.7	0.5	0.0	0.0	0.0	1.4	1.3	0.6	1.0	1.1	1.0	0.8	0.9
C10:0	2.4	0.6	0.1	0.1	0.0	3.1	3.2	1.4	2.4	2.6	2.5	2.1	2.2
C12:0	2.1	0.9	0.3	0.2	0.4	3.6	3.8	1.6	3.1	3.2	2.9	2.7	2.7
C14:0	6.7	3.7	1.7	0.9	1.8	12.9	12.5	5.8	10.8	10.6	9.9	9.3	9.0
C16:0	28.6	30.7	26.7	25.4	31.6	40.3	40.5	30.9	30.8	30.6	30.0	30.3	29.5
C18:0	5.8	4.8	2.5	2.2	2.7	13.3	7.1	6.3	9.2	8.4	8.1	8.1	7.5
C18:1 n-9 cis	16.5	17.0	13.5	15.8	15.1	10.6	12.7	18.8	21.1	20.1	19.6	19.8	19.1
C18:2 n-6 cis	22.5	32.8	48.8	49.6	42.9	3.8	9.4	24.3	11.5	13.3	16.0	18.1	19.9
C18:3 n-3	0.0	0.0	0.0	0.0	0.0	0.1	0.1	0.5	1.2	1.3	1.4	1.4	1.5
Total saturates	55.2	45.1	31.5	29.0	36.5	78.3	71	47.9	59.4	58.8	57.0	54.8	53.6
Ratio TU[1]/TS[2]	0.71	1.10	1.98	2.25	1.59	0.18	0.31	0.91	0.57	0.59	0.65	0.72	0.76

[1]TU: Total unsaturated fatty acids; [2]TS: Total saturated fatty acids.
The presented data are the means of three replicate samples for different retail brands of tarhana (T1-T5) or kishk (K1-K3) powders and different experimental FMB consisting of 20-40% bulgur and 80-60% fermented milk (FMB80-FMB60)

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Table 4

β-Glucan and phytic acid content of tarhana, kishk powders and experimental fermented milk:bulgur blends (FMB).

	β-Glucan	Phytic acid
Tarhana		
T1	127.2±5.6$^{b\text{-}e,g,h}$	132.5±46.0$^{f\text{-}h}$
T2	23.1±1.2$^{a,c,f\text{-}h}$	55.8±13.6$^{f\text{-}h}$
T3	55.1±13.8$^{a,b,f\text{-}h}$	69.1±17.5$^{f\text{-}h}$
T4	38.2±5.7$^{a,f\text{-}h}$	130.3±12.0$^{f\text{-}h}$
T5	49.7±2.9$^{a,f\text{-}h}$	102.6±5.2$^{f\text{-}h}$
Kishk		
K1	113.3±9.9$^{b\text{-}e,f,h}$	680.6±68.5$^{a\text{-}e,h}$
K2	241.2±17.3$^{a\text{-}f,h}$	598.8±75.7$^{a\text{-}e,h}$
K3	344.8±16.4$^{a\text{-}g}$	426.7±54.0$^{a\text{-}g}$
Experimental FMB		
FMB80	169.2±6.3$^{C\text{-}E}$	369.9±50.4
FMB75	184.1±15.2^{D,E}	456.3±60.1
FMB70	218.4±26.6^{A}	403.8±46.3
FMB65	238.7±21.4^{A,B}	443.2±44.8
FMB60	229.3±14.6^{A,B}	465.4±53.9

The presented data are the means of three replicate samples for different retail brands of tarhana (T1-T5) or kishk (K1-K3) powders and different experimental FMB consisting of 20-40% bulgur and 80-60% fermented milk (FMB80-FMB60)

$^{a\text{-}h}$ indicates significant difference (P<0.05, ANOVA followed by Tukey's test) between kishk or tarhana samples (^{a} is T1, ^{b} is T2 etc.)

$^{A\text{-}E}$ indicates significant difference (P<0.05, ANOVA followed by Tukey's test) between experimental FMB samples (^{A} is T1, ^{B} is T2 etc.)

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Table 5

Antioxidant activity of retail brands of tarhana and kishk and experimental fermented milk:bulgur blends (FMB).

	TPC[1] (mg GAE/100g)	FRAP[2] (mmol Fe^{2+}/100g)	ORAC[3] (mg TE/100g)
Tarhana			
T1	64.4±2.9^{c-e}	62.9±14.0^{c-e}	28.1±2.7^{d,e}
T2	71.7±7.4^{c-e}	72.9±8.8^{c-e}	26.5±3.3^{d,e}
T3	127.8±13.5$^{a,b,d,f-h}$	224.8±49.2$^{a,b,e-h}$	34.4±4.2$^{d,f-h}$
T4	179.7±13.7$^{a-c,e-h}$	219.8±40.7$^{a,b,e-h}$	44.9±2.9$^{a-c,f-h}$
T5	141.2±5.8$^{a,b,d,f-h}$	357.6±40.0$^{a-d,f-h}$	37.9±5.5$^{a,b,f-h}$
Kishk			
K1	62.1±3.9^{c-e}	137.3±22.7^{c-e}	23.8±3.6^{c-e}
K2	56.4±5.2^{c-e}	111.1±25.2^{c-e}	21.4±1.8^{c-e}
K3	47.3±3.4^{c-e}	123.6±9.0^{c-e}	19.5±1.4^{c-e}
Experimental FMB			
FMB80	52.5±4.7^{D}	66.0±5.1	23.3±0.7
FMB75	48.0±3.2	61.6±5.3	23.9±1.3
FMB70	45.0±5.0	61.9±5.7	23.5±1.1
FMB65	41.2±3.5^{A}	58.1±3.8	25.4±1.6
FMB60	46.5±4.4	61.3±6.2	25.9±1.3

[1]TPC, total phenolic content expressed as gallic acid equivalents (GAE)/100g; [2]FRAP, ferric reducing antioxidant power expressed as mmol Fe^{2+}/100g; [3]ORAC, oxygen radical absorbance capacity expressed as trolox equivalents (TE)/100g.

The presented data are the means of three replicate samples for different retail brands of tarhana (T1-T5) or kishk (K1-K3) powders and experimental FMB consisting of 20-40% bulgur and 80-60% fermented milk(FMB80-FMB60)

$^{a-h}$ indicates significant difference (P<0.05, ANOVA followed by Tukey's test) between kishk or tarhana samples (^{a} is T1, ^{b} is T2 etc.)

$^{A-E}$ indicates significant difference (P<0.05, ANOVA followed by Tukey's test) between experimental FMB samples (^{A} is T1, ^{B} is T2 etc.)

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Table 6-Details of retail brands of Tarhana and Kishk powders

Brand	Origin	Ingredients
Tarhana		
T1	Greece	durum wheat semolina, yoghurt, milk butter, salt
T2	Turkey	wheat flour, yoghurt, salt, red pepper, tomato, onion, peas, dry beans, lentils
T3	Turkey	wheat flour, yoghurt, salt, red pepper, onion, mint, sour yeast
T4	Turkey	wheat flour, yoghurt, red pepper, onion, mint
T5	Turkey	wheat flour, yoghurt, salt, red pepper, tomato, onion, sour yeast
Kishk		
K1	Lebanon	bulgur wheat, yoghurt, salt
K2	Lebanon	bulgur wheat, yoghurt, salt
K3	Syria	bulgur wheat, yoghurt, salt

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